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Research Article

Small reserve but high diversity: butterfly community across an altitudinal gradient in the Brazilian Atlantic Forest

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Abstract

The present study aimed to describe the composition of the butterfly community in relation to the altitudinal gradient in the Reserva Particular do Patrimônio Natural (RPPN) Alto-Montana, Serra da Mantiqueira, Minas Gerais, Brazil, and to provide a list of species for the area. We collected samples in the RPPN Alto-Montana along an altitudinal gradient from 1400 to 2100 m, between the dry and rainy seasons of 2018 and 2019. During this period, the sampling method utilizing Van-Someren Rydon traps totaled 3,936 hours and the effort using sweep nets totaled 246 hours. A total of 1,253 butterflies distributed across 124 species and six families of diurnal butterflies were observed (Hesperiidae, Lycaenidae, Nymphalidae, Papilionidae, Pieridae, and Riodinidae). Nymphalidae was the most representative family, followed by Hesperiidae, Pieridade, Lycaenidae, Papilionidae and Riodinidae. Moreover, we recorded 37 species with only one individual (*singletons*) and 20 species with only two individuals (*doubletons*), totalling 57 species, which corresponds to 46% of all sampled richness. The rarefaction curve did not reveal a tendency toward stabilization. However, the indices showed slightly higher values for the 124 species sampled. The analysis performed using the Bootstrap estimator predicted a total of 143.22 species (± 10.87 SE), with a further 19 additional species than observed. Chao 1 predicted 153.42 (± 11.82 SE), and Jackknife 1 predicted 164.00 (± 16.29 SE) species, with 29 and 40 additional species than the observed, respectively. Our study contributes to the knowledge of butterfly biodiversity in Serra da Mantiqueira and reveals a high species richness for the RPPN Alto Montana, especially considering the relatively small area. In addition, our study provides the first inventory of butterflies for the RPPN Alto Montana, thus supporting further studies investigating the butterfly richness in the Serra da Mantiqueira region. Finally, our findings of endemic, rare, and endangered butterfly species highlight the relevance of further conservation strategies to be considered for the Protected Area's Management Plan.

Key words: Conservation, Hesperiidae, inventory, Lepidoptera, Nymphalidae, species richness, threatened species

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Introduction

Covering part of the Atlantic coast of South America, the Atlantic Forest Domain is composed of a mosaic of tropical vegetation with typical forest formations, altitude grasslands, restingas, and mangroves (Joly et al. 2014; Soldati et al. 2019; Marques and Grelle 2021). With the urbanization of recent centuries, resource exploitation, and land-use change, much of the Atlantic Forest's cover has been deforested and fragmented, with today only around 11% of its original area remaining (Ribeiro et al. 2009; de Lima 2020). Approximately 90% of its extension is located in Brazilian territory (Ribeiro et al. 2009; Colli-Silva et al. 2020), and its great richness of fauna and flora associated with high rates of endemism make this Domain a biodiversity hotspot (Myers et al. 2000). This richness is reflected in the fact that the Atlantic Forest is home to an enormous diversity of Lepidoptera. One of the reasons for this diversity is the vegetation associated with altitudinal and climatic variations, which provides heterogeneous environments that favor the occurrence of several species of butterflies (Soldati et al. 2019).

The butterfly richness for the Atlantic Domain is approximately 2,100 species (Francini et al. 2011). These values correspond to about 59% of the total butterfly richness for Brazil - 3,517 species (Casagrande and Duarte 2023) and 23% of the total butterfly richness for Latin America - 9,000 species (Raven et al. 2020). This high diversity can be explained by environmental characteristics such as habitat heterogeneity, and also by inter and intra specific interactions such as competition, factors that reflect on the different colors, body shape, and diet of these organisms in their different life stages - juvenile or adult (Stevens et al. 2012). Different life stages allow butterflies to occupy a wide range of habitats, mainly because they do not compete for food. When in the juvenile stage, they have mandibles and feed on leaves, flowers, and other plant parts such as stems (Stevens et al. 2012). When adults, they have proboscis and are classified into two major guilds: Fruit-feeding when food is based on fermented fruits, sap, and/or exudates, and nectarivores when flower nectar is their food resource (DeVries et al. 1997). Butterflies are also involved in various ecological interactions such as pollination, herbivory, plant population dynamics, and serving as food for higher trophic level organisms (DeVries et al. 1997).

All these characteristics classify them as bioindicators of environmental quality and illustrate the capacity with which this group responds quickly to landscape and climate changes (Ribeiro et al. 2012). Thus, butterflies are an excellent study group because they are sensitive to environmental disturbances, allowing to better evaluate the impacts caused to the environment (DeVries et al. 1997). In addition, they are organisms that are relatively easy to capture, having short life cycles and well-known taxonomy (Ribeiro et al. 2012). That way, even though they are an abundant group, butterflies have been suffering major population declines (Sánchez-Bayo and Wyckhuys 2019). This decline comes from the increasing loss of habitat caused by the exploitation of natural resources, deforestation, intensive agriculture, pollution, climate change, and species introduction (Sánchez-Bayo and Wyckhuys 2019). Today, many species are listed as endangered species (Chowdhury et al. 2023). Given this scenario, studies aimed at understanding patterns of distribution and diversity of butterflies are important tools to better support conservation actions, such

as the creation of conservation units and improved wildlife management (Chowdhury et al. 2023).

Although studies with butterflies have increased in recent decades, inventories for this group in mountainous regions are still scarce (Quintero and Jetz 2018; Girardello et al. 2019; Shirai et al. 2019). Some explanations for this scarcity are these places' logistical and natural difficulties, such as rugged terrain and access difficulties (Quintero and Jetz 2018; Girardello et al. 2019). In the Serra da Mantiqueira region, some efforts aimed at understanding the composition of the butterfly community were carried out by Zikán and Zikán (1968), Freitas et al. (2011) and Vieira et al. (2022). This study aims to complement the knowledge about the butterfly community of Serra da Mantiqueira, particularly for the southern region of Minas Gerais. Therefore, our objective is to describe the composition of the butterfly community related to the altitudinal gradient in the Reserva Particular do Patrimônio Natural (RPPN) Alto-Montana, Serra da Mantiqueira, Minas Gerais, Brazil, and to provide a list of species for the area.

Materials and methods

Study area

The study was carried out in the Reserva Particular do Patrimônio Natural (RPPN) Alto-Montana, in the municipality of Itamonte, in the southern region of the state of Minas Gerais, Brazil (22°21'55"S, 44°48'32"W, Fig. 1). The reserve has about 672 ha and is part of the Serra da Mantiqueira Environmental Protection Area (Mazza et al. 2018; Souza et al. 2021). The landscape is quite rugged, with a granite outcrop that stands out in the landscape, ranging from 1,400 to 2,500 m in altitude. Its climate is subtropical highland climate (Cwb), with mild, dry winters and rainy summers (Alvares et al. 2013). The average annual temperature is 18.6 °C, and the average rainfall is 1,749 mm, with periods of heavy rain from December to January (Cruz et al. 2014; Vilanova 2015; ICMBio 2018). The vegetation is diverse, being composed of typical phytophysiognomies of the Atlantic Domain, with the presence of Floresta Estacional Semidecidual, Floresta Ombrófila Densa Montana, Floresta Ombrófila Densa Altomontana (Cloud Forest), Floresta Ombrófila Mista and Campos de Altitude in the higher altitudes (Fig. 2A, B). The RPPN Alto Montana is one of the only places in Minas Gerais with the native presence of *Araucaria angustifolia* (Bertol.) Kuntze, the dominant species of Floresta Ombrófila Mista (Cruz et al. 2014; Pompeu et al. 2014; ICMBio 2018).

Data sampling

The butterfly samplings were carried out along the altitudinal gradient in an access trail in the northern area of the RPPN Alto-Montana. This region of the protected area has a slope of 700 meters starting from 1,400 m to a maximum altitude of 2,100 m above sea level. In order to carry out the sampling of butterflies along the gradient, eight altitudinal elevations were delimited with the aid of a Garmin® GPS, distributed across every 100 m of altitude, over a period of one year, in four collection periods, contemplating the rainy season, the dry season, and the transition between the two. Sampling in different seasons ensures that species that have life cycles in different periods can be captured and sampled,

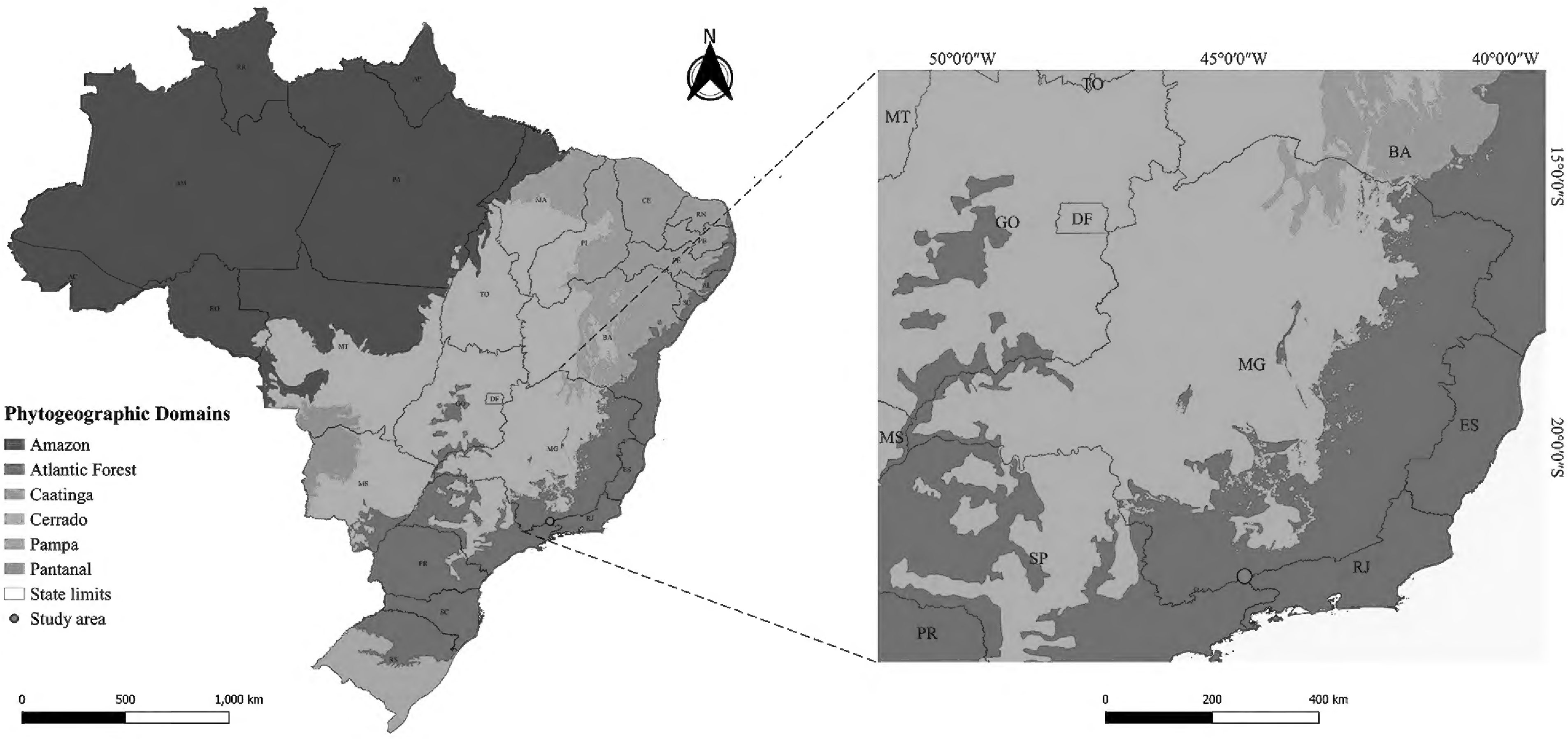

Figure 1. Map of the geographic location of the study area in Reserva Particular do Patrimônio Natural (RPPN) Alto-Montana, Itamonte, Minas Gerais, Brazil. The boundaries of Brazilian phytogeographic domains were adapted from shapefiles available from the IBGE - Instituto Brasileiro de Geografia e Estatística (2023), from the global ecoregions of Dinerstein et al. (2017), and from the map of vegetations on rocky outcrops of the Cerrado domain by Newton Barbosa. Map design: Cássio Cardoso Pereira.

thus generating more robust results (Ribeiro et al. 2016). Therefore, to contemplate the transition between the rainy season of 2017 and the dry season of 2018, the samplings were carried out in February 2018 (rainy season, three days) and April 2018 (dry season, three days). Subsequently, we carried out samplings in July 2018 (dry season, four days) and January 2019 (rainy season, three days). In this way, we totaled thirteen days of collection throughout the evaluated period.

For the capture of frugivorous butterflies, 40 traps of the Van-Someren Rydon type were distributed in groups of five traps for each altitudinal level (Fig. 2C, D), separated by at least 20 m and placed at a height of approximately 1 m from the ground, in all altitude ranges. The traps contained a bait made out of a mixture of banana and sugarcane molasses that underwent 48 hours of fermentation and were left on site for 48 hours (e.g., Uehara-Prado et al. 2009; Melo et al. 2019). During this period, the traps were inspected after 24 hours to remove captured individuals (Uehara-Prado et al. 2009; Santos et al. 2011; Henriques et al. 2019).

The collection and active search for butterflies along the gradient were carried out via sweep nets in each altitudinal range, during the same period of 1 year, to capture nectarivorous or even frugivorous butterflies that were eventually flying or foraging. The time for these samplings comprised an interval between 9 am and 4 pm when the butterflies are more active and thus more susceptible to active capture. The collections lasted around 50 minutes for each of the eight gradients, alternating the starting points, that is, on one day we started at 1400 m at 9 am and finished at 2100 m at 4 pm. The next day, we started at 2100 m at 9 am and ended at 1400 m at 4 pm (Santos et al. 2011; Henriques et al. 2019). All individuals sampled were placed in entomological envelopes with records of altitude, date, and place of collection. Subsequently, they were sacrificed, mounted, and identified via guides and/or consultations with entomological collections and specialists. All species were checked for

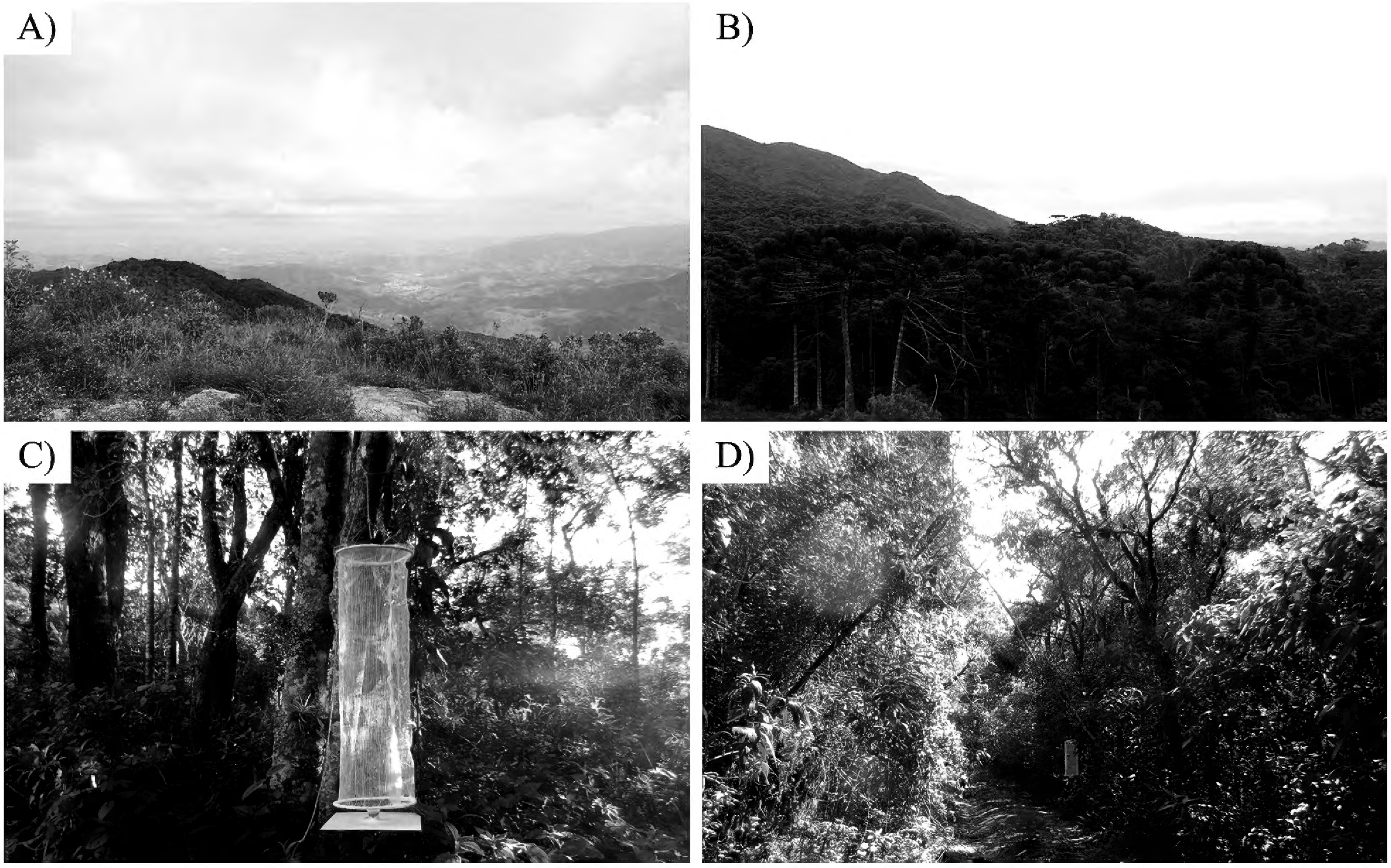

Figure 2. Sampling areas of the Reserva Particular do Patrimônio Natural (RPPN) Alto-Montana, Itamonte, Minas Gerais, Brazil **A** Campo de Altitude **B** Floresta Ombrófila Mista **C, D** Van-Someren Rydon type trap installed.

their conservation status by consulting the International Union for Conservation of Nature – IUCN website (2023). The taxonomy adopted for this study is according to the database of the Global Biodiversity Information – GBIF (2022). The reference collection was organized and deposited in the Entomological Collection of the Departamento de Ciências Naturais of the Universidade Federal de Minas Gerais (UFMG).

The sampling effort was calculated based on the hours of active collections and baited traps. The sampling effort per baited trap was obtained by multiplying the total number of traps placed in the field for all collection days and the total number of hours the traps were available to butterflies on selected sites. The sampling effort per active collection was calculated by multiplying the number of hours (for each collection day) by the number of sweep nets used. Finally, the total sampling effort was obtained by adding the efforts by active collection and baited trap of each day sampled. Thirteen field trips were carried out in the (RPPN) Alto-Montana. During this period, the sampling effort by traps totaled 3,936 hours and the effort by sweep nets totaled 246 hours.

Data analysis

The total richness was estimated for the entire length of the altitudinal gradient. All species were analyzed and classified according to their frequency of occurrence in order to detect rare species. Species in which only one individual was observed were classified as *singletons*. Species with two individuals observed were classified as *doubletons*, and species with more than two individuals were considered common species (Chao 1984; Ferraz et al. 2009; Colwell 2013).

To assess sampling sufficiency, a rarefaction curve was constructed using the *specaccum* function in Vegan (Oksanen 2013) in R software (R Core Team 2021). The *specaccum* function acts by randomizing the sample and describing the average species accumulation curve (plus standard deviation) from the application of a thousand permutations of the data, eliminating the temporal bias (Oksanen 2013). The rarefaction method was based on the 13 sampling periods. Using the specpool function in Vegan (Oksanen 2013), three richness estimator indices (Bootstrap, Chao 1, Jackknife 1) were calculated. Jackknife 1 is an index based on the number of species that occur only once in the original sample (*singletons*), being an analysis sensitive to rare species (Quenouille 1956; Ferraz et al. 2009; Colwell 2013). Chao 1, like Jackknife 1, is also a sensitive estimator for rare species (Chao 1984; Chao and Shen 2004; Colwell et al. 2012). However, in Chao 1 the estimated richness takes into account not only the number of species represented by a single individual (*singletons*) but also the number of species with an abundance of two individuals (*doubletons*) (Chao 1984; Ferraz et al. 2009). Finally, Bootstrap uses data from all collected species to estimate total richness without giving greater weight to species that appear less often in the sample (Efron 1979; Smith and Van Belle 1984; Colwell 2013).

Results

A total of 1,253 butterflies distributed in 124 species and six families of diurnal butterflies (Hesperiidae, Lycaenidae, Nymphalidae, Papilionidae, Pieridae, and Riodinidae) were observed (Table 1, see some of these species in Figs 3–5). Nymphalidae (65 species, about 52.42% of the total) was the most representative family, followed by Hesperiidae (27 species, 21.77%), Pieridade (14 species, 11.29%), Lycaenidae (8 species, 6.45%), Papilionidae (6 species, 4.84%) and Riodinidae (4 species, 3.23%). Nymphalidae was also the most abundant family with 841 individuals sampled, corresponding to 67.12% of the total abundance. Pieridae was the second most abundant family (230 individuals, 18.36%), followed by Hesperiidae (126 individuals, 10.05%), Papilionidae (37 individuals, 2.95%) Lycaenidae (11 individuals, 0.88%), and Riodinidae (8 individuals, 0.64%).

The most abundant species were *Morpho portis* (Hübner, 1821) (Nymphalidae: Satyrinae, N = 249, Fig. 4F) followed by *Dismorphia thermesia* (Godart, 1819) (Pieridae: Dismorphiinae, N = 128, Fig. 3F), *Foetterleia schreineri* (Foetterle, 1902) (Nymphalidae: Satyrinae, N = 85, Fig. 3I), *Forsterinaria necys* (Godart, 1824) (Nymphalidae: Satyrinae, N = 78), *Heliconius besckei* (Ménétriés, 1857) (Nymphalidae: Heliconiinae, N = 62, Fig. 4A), *Eurema albula* (Cramer, 1775) (Pieridae: Coliadinae, N = 58), and *Praepedaliodes phanias* (Hewitson, 1861) (Nymphalidae: Satyrinae, N = 40). Together, these species account for about 56% of the total abundance. Of these, only *M. portis* was recorded in a single season of the year (rainy season - January 2019), the other species occurred in all seasons. Considering the altitudinal gradient, only *M. portis* and *F. necys* occurred at all altitudinal points. Thirty-seven species with only one individual (*singletons*) and 20 species with only two individuals (*doubletons*) were recorded, totaling 57 species, which corresponds to 46% of all sampled richness.

Table 1. List of butterfly species from Reserva Particular do Patrimônio Natural (RPPN) Alto-Montana, Itamonte, Minas Gerais states, Brazil. S = richness; m = meters; n = number of individuals. IUCN Status: NE = Not Evaluated; LC = Least Concern.

Family/Species	Altitudinal gradient (m)/Abundance (n)								Total	IUCN Status
	1400	1500	1600	1700	1800	1900	2000	2100		
Hesperiidae (S = 27)										
Hesperiinae (S = 15)										
Callimormus saturnus (Herrich-Schäffer, 1869)	–	–	–	1	–	–	–	–	1	NE
Calpodes ethlius (Stoll, 1782)	–	2	–	2	3	5	3	–	15	NE
Cobalopsis nero (Herrich-Schäffer, 1869)	–	1	–	–	–	–	–	–	1	NE
Cumbre cumbre (Schaus, 1902)	–	–	–	–	–	3	–	–	3	NE
Cymaenes gisca (Evans, 1955)	2	1	–	2	7	5	8	4	29	NE
Cymaenes lepta (Hayward, 1938)	–	–	–	–	–	–	–	1	1	NE
Cymaenes tripunctata (Latreille, 1824)	–	–	–	–	1	–	–	–	1	NE
Cymaenes tripunctus (Herrich-Schäffer, 1865)	1	1	–	1	6	1	1	–	11	NE
Lucida lucia (Capronnier, 1874)	–	1	–	–	–	–	–	–	1	NE
Vehilius clavicula (Plötz, 1884)	–	–	–	–	2	–	–	–	2	NE
Vettius aurelius (Plötz, 1882)	–	–	1	2	1	–	–	–	4	NE
Vettius diversa (Herrich-Schäffer, 1869)	–	–	1	–	–	–	–	–	1	NE
Vettius phyllus (Cramer, 1777)	–	–	–	1	1	–	–	–	2	NE
Vettius ploetzii (Capronnier, 1874)	–	–	1	–	6	5	1	–	13	NE
Zariaspes mys (Hübner, 1808)	–	–	–	1	–	–	–	–	1	NE
Eudaminae (S = 1)										
Autochton neis (Geyer, 1832)	–	–	–	–	–	1	–	–	1	NE
Pyrginae (S = 11)										
Achlyodes busirus (Stoll, 1782)	–	–	–	–	–	1	2	1	4	NE
Astraptes fulgerator (Walchs, 1775)	1	1	1	–	–	–	–	–	3	NE
Epargyreus socus (Hübner, 1925)	–	1	1	–	–	–	–	–	2	NE
Heliopetes ochroleuca (Zikán, 1938)	2	3	1	–	1	1	–	–	8	NE
Oechydrus chersis (Herrich-Schäffer, 1869)	5	1	–	–	–	–	–	–	6	NE
Pyrgus orcus (Stoll, 1780)	–	–	–	–	–	1	–	–	1	NE
Pythonides lancea (Hewitson, 1868)	1	1	–	–	–	–	–	–	2	NE
Sostrata cronion (C. Felder & R. Felder, 1867)	2	–	–	–	–	–	–	–	2	NE
Theagenes dichrous (Mabille, 1878)	–	–	–	–	–	–	–	1	1	NE
Urbanus dorantes (Stoll, 1790)	–	–	–	–	1	–	–	–	1	NE
Urbanus teleus (Hübner, 1821)	1	2	4	–	1	–	–	1	9	NE
Lycaenidae (S = 8)										
Theclinae (S = 8)										
Arawacus meliboeus (Fabricius, 1793)	–	–	–	–	–	1	–	–	1	NE
Arawacus tadita (Hewitson, 1877)	–	–	–	–	–	1	–	–	1	NE
Laothus phydela (Hewitson, 1867)	–	–	–	1	–	–	–	–	1	NE
Strymon bubastus (Stoll, 1780)	–	–	–	1	–	1	–	–	2	NE
Symbiopsis lenitas (Druce, 1907)	–	–	1	–	1	–	1	–	3	NE
Theritas deniva (Hewitson, 1874)	–	–	–	–	–	1	–	–	1	NE
Theritas triquetra (Hewitson, 1865)	–	–	–	–	–	–	–	1	1	NE
Thestius azaria (Hewitson, 1867)	–	–	–	–	–	–	–	1	1	NE

Family/Species	Altitudinal gradient (m)/Abundance (n)								Total	IUCN Status
	1400	1500	1600	1700	1800	1900	2000	2100		
Nymphalidae (S = 65)										
Biblidinae (S = 4)										
Catonephele sabrina (Hewitson, 1852)	1	–	2	–	6	2	–	1	12	NE
Diaethria candrena (Godart, 1823)	2	–	–	–	–	1	–	–	3	NE
Epiphile hubneri Hewitson, 1861	1	4	13	2	3	–	–	–	23	NE
Epiphile orea (Hübner, 1823)	–	4	8	7	1	6	–	–	26	NE
Charaxinae (S = 4)										
Archaeoprepona chalciope (Hübner, 1823)	1	1	–	–	–	–	–	–	2	NE
Memphis moruus (Fabricius, 1775)	–	1	2	–	–	–	–	–	3	NE
Polygrapha suprema (Schaus, 1920)	–	–	–	–	4	5	3	5	17	NE
Zaretis itys (Cramer, 1777)	–	–	2	–	–	–	–	–	2	NE
Danainae (S = 7)										
Episcada carcinia (Schaus, 1902)	–	–	–	1	–	–	–	–	1	NE
Episcada philoclea (Hewitson, 1854)	–	1	–	–	–	–	–	1	2	NE
Epityches eupompe (Geyer, 1832)	–	–	2	–	1	–	1	–	4	NE
Hypothyris ninonia (Hübner, 1806)	1	1	1	–	–	–	–	–	3	NE
Mechanitis lysimnia (Fabricius, 1793)	1	–	3	–	1	–	–	–	5	NE
Pseudoscada erruca (Hewitson, 1855)	2	–	1	–	–	–	–	–	3	NE
Pteronymia sylvo (Geyer, 1832)	1	–	1	–	4	–	–	–	6	NE
Heliconiinae (S = 12)										
Actinote alalia (Felder, 1860)	–	–	–	–	1	1	–	2	4	NE
Actinote bonita (Penz, 1996)	–	–	–	–	–	3	–	–	3	NE
Actinote conspicua (Jordan, 1913)	–	–	–	–	1	–	–	2	3	NE
Actinote dalmeidai (Francini, 1996)	–	–	–	–	–	–	–	1	1	NE
Actinote mantiqueira (Freitas, Francini, Paluch & Barbosa, 2018)	–	–	1	–	2	–	2	6	11	NE
Actinote surima (Schaus, 1902)	–	–	–	–	–	–	–	3	3	NE
Dione juno (Cramer, 1779)	–	–	–	–	–	1	–	–	1	NE
Eueides pavana (Ménétriés, 1857)	2	–	–	–	–	–	–	–	2	NE
Heliconius besckei (Ménétriés, 1857)	–	–	2	1	1	–	–	–	4	NE
Heliconius erato (Linnaeus, 1758)	11	9	11	12	14	4	1	–	62	NE
Heliconius ethilla (Latreille & Godart, 1819)	2	4	5	1	–	1	–	–	13	NE
Philaethria wernickei (Röber, 1906)	–	–	1	–	–	–	–	–	1	NE
Limenitidinae (S = 6)										
Adelpha calliphane (Fruhstorfer, 1915)	3	2	1	–	–	–	–	–	6	NE
Adelpha hyas (Boisduval, 1836)	–	–	–	–	–	–	–	1	1	NE
Adelpha mythra (Godart, 1823)	1	2	4	3	–	–	–	–	10	NE
Adelpha poltius (Hall, 1938)	–	–	–	1	2	–	1	–	4	NE
Adelpha syma (Godart, 1823)	–	–	–	–	–	–	1	1	2	NE
Adelpha thessalia (Felder, 1867)	–	–	–	1	–	–	–	–	1	NE
Nymphalinae (S = 4)										
Eresia lansdorfi (Godart, 1819)	–	–	1	–	–	–	–	–	1	NE
Tegosa anieta (Hewitson, 1864)	1	3	–	2	2	–	1	–	9	NE
Tegosa claudina (Eschscholtz, 1821)	6	3	4	–	–	1	–	–	14	NE
Vanessa braziliensis (Moore, 1883)	–	–	–	–	–	1	–	2	3	NE
Satyrinae (S = 28)										
Caligo arisbe (Hübner, 1822)	–	–	1	–	–	–	–	–	1	NE
Caeruleuptychia helena (Anken, 1994)	1	–	1	–	–	–	–	–	2	NE

Family/Species	Altitudinal gradient (m)/Abundance (n)								Total	IUCN Status
	1400	1500	1600	1700	1800	1900	2000	2100		
Carminda griseldis (Weymer, 1911)	–	–	–	1	–	–	1	–	2	NE
Carminda surpresa (Barbosa, Aguiar, Rosa, Zacca & Freitas, 2020)	1	–	–	–	–	–	–	–	1	NE
Carminda umuarama (Ebert & Dias, 1997)	1	–	–	1	–	1	8	7	18	NE
Cissia phronius (Godart, 1824)	–	–	1	1	2	3	–	–	7	NE
Eryphanis reevesii (Doubleday, 1849)	–	–	1	–	–	–	–	–	1	NE
Eteona tisiphone (Boisduval, 1836)	1	–	–	–	–	–	1	–	2	NE
Foetterleia schreineri (Foetterle, 1902)	1	2	–	–	16	23	18	25	85	NE
Forsterinaria necys (Godart, 1824)	5	2	9	6	22	21	10	3	78	NE
Forsterinaria pronophila (Butler, 1867)	2	–	1	1	–	–	1	1	6	NE
Forsterinaria quantius (Godart, 1824)	15	–	–	–	–	–	–	–	15	NE
Godartiana muscosa (Butler, 1870)	1	–	3	3	1	–	–	–	8	NE
Moneuptychia itapeva (Freitas, 2007)	–	–	–	–	–	–	–	13	13	NE
Moneuptychia montana (Freitas, 2015)	–	–	–	–	–	–	3	2	5	NE
Moneuptychia pervagata (Freitas, Siewert & Mielke, 2015)	1	–	–	–	1	1	–	2	5	NE
Morpho epistrophus (Fabricius, 1796)	7	–	–	–	–	–	–	–	7	NE
Morpho portis (Hübner, 1821)	5	20	38	40	71	39	27	9	249	NE
Narope cyllastros (Doubleday, 1849)	–	–	–	–	2	–	1	–	3	NE
Opoptera syme (Hübner, 1821)	–	–	1	1	–	–	–	–	2	NE
Paryphthimoides eous (Butler, 1866)	–	–	1	–	–	–	1	1	3	NE
Paryphthimoides poltys (Prittwitz, 1865)	1	–	–	1	–	–	–	–	2	NE
Praepedaliodes phanias (Hewitson, 1861)	–	–	–	2	7	11	18	2	40	NE
Pseudodebis ypthima (Hübner, 1821)	–	–	–	–	–	1	–	–	1	NE
Splendeuptychia libitina (Butler, 1870)	–	1	1	2	4	–	–	1	9	NE
Taygetis ypthima (Hübner, 1816)	–	–	1	–	–	–	–	–	1	NE
Yphthimoides angularis (Butler, 1867)	–	2	–	–	1	–	–	–	3	NE
Yphthimoides ochracea (Butler, 1867)	–	–	–	–	–	–	–	1	1	NE
Papilionidae (S = 6)										
Papilioninae (S = 6)										NE
Heraclides hectorides (Esper, 1794)	2	1	1	–	–	–	–	–	4	LC
Mimoides lysithous (Hübner, 1821)	4	–	–	–	–	–	–	–	4	LC
Parides agavus (Drury, 1782)	1	1	–	–	–	–	–	–	2	LC
Parides anchises (Linnaeus, 1758)	1	–	–	–	–	–	–	–	1	LC
Parides bunichus (Hübner, 1821)	6	9	–	–	–	–	–	–	15	LC
Parides proneus (Hübner, 1831)	4	6	–	1	–	–	–	–	11	LC
Pieridae (S = 14)										
Coliadinae (S = 6)										
Eurema albula (Cramer, 1775)	8	16	30	2	1	1	–	–	58	NE
Eurema deva (Doubleday, 1847)	1	–	2	–	–	1	–	1	5	NE
Eurema elathea (Cramer, 1777)	3	–	–	–	–	–	–	–	3	NE
Eurema lirina (Bates, 1861)	3	3	–	1	–	–	–	–	7	NE
Eurema phiale (Cramer, 1775)	–	–	–	–	–	–	–	1	1	NE
Pyrisitia nise (Cramer, 1775)	–	–	–	–	1	–	–	–	1	NE
Dismorphiinae (S = 3)										
Dismorphia amphione (Cramer, 1779)	–	1	–	1	–	–	–	–	2	NE
Dismorphia astyocha (Hübner, 1831)	–	–	1	–	–	1	–	–	2	NE

Family/Species	Altitudinal gradient (m)/Abundance (n)								Total	IUCN Status
	1400	1500	1600	1700	1800	1900	2000	2100		
Dismorphia thermesia (Godart, 1819)	6	23	24	40	32	2	1	–	128	NE
Pierinae (S = 5)										
Archonias brassolis (Fabricius, 1776)	–	1	–	–	–	–	–	–	1	NE
Catasticta bithys (Hübner, 1831)	–	–	–	–	5	1	2	1	9	NE
Hesperocharis anguitia (Godart, 1819)	–	–	–	–	–	–	–	1	1	NE
Hesperocharis erota (Lucas, 1852)	–	–	–	–	1	1	5	3	10	NE
Pereute swainsoni (Gray, 1832)	–	1	–	–	–	–	1	–	2	NE
Riodinidae (S = 4)										
Riodininae (S = 4)	–	–	–	–	–	–	–	–		
Charis cadytis (Hewitson, 1866)	–	1	1	1	–	–	–	–	3	NE
Mesosemia rhodia (Godart, 1824)	–	–	–	1	2	–	–	–	3	NE
Synargis paulistina (Stichel, 1910)	–	–	–	–	–	–	1	–	1	NE
Synargis regulus (Fabricius, 1793)	1	–	–	–	–	–	–	–	1	NE

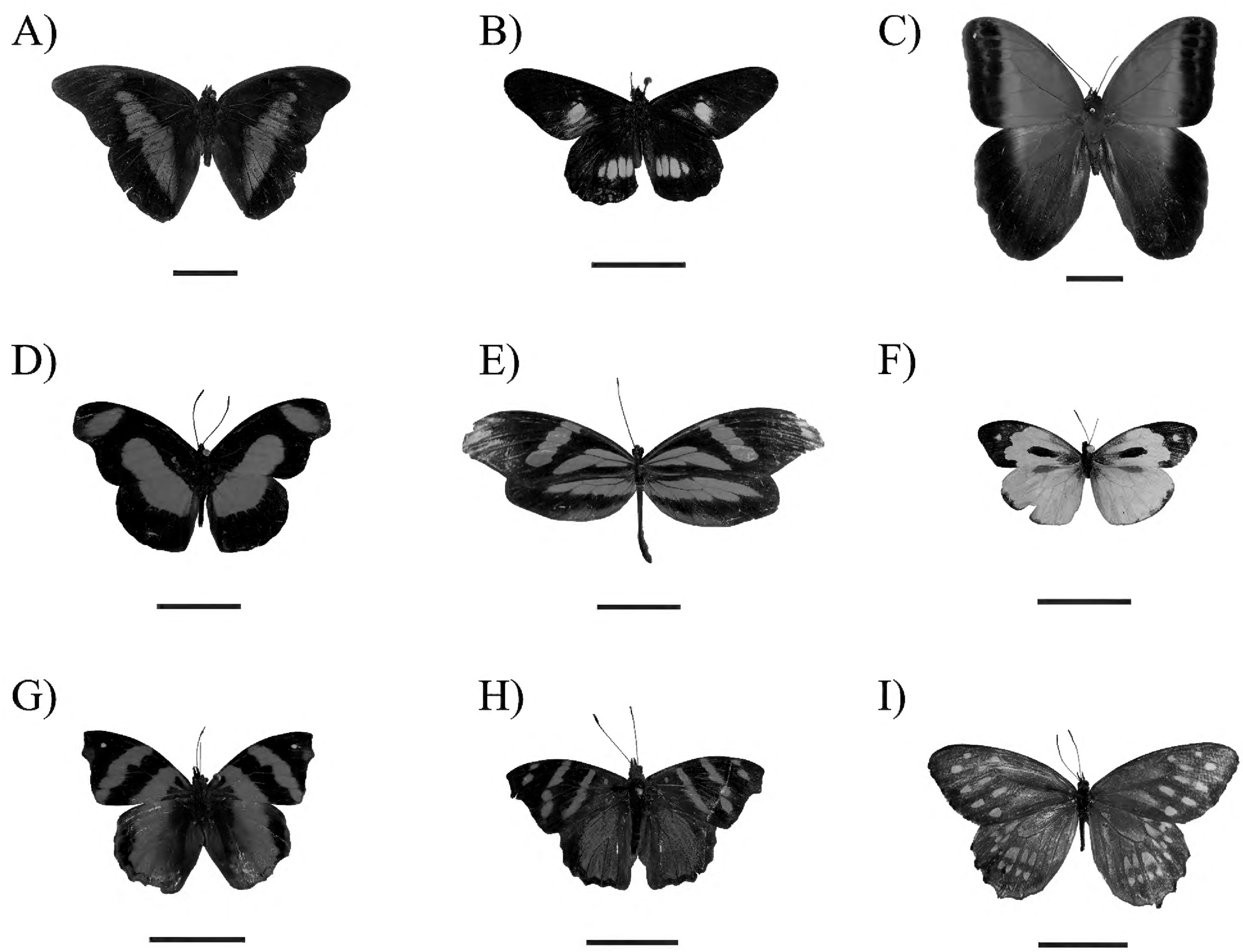

Figure 3. Butterfly species recorded in Reserva Particular do Patrimônio Natural (RPPN) Alto-Montana, Itamonte, Minas Gerais, Brazil **A** *Archaeoprepona chalciope* (Hübner, 1823) **B** *Archonias brassolis* (Fabricius, 1776) **C** *Caligo arisbe* (Hübner, 1922) **D** *Catonephele sabrina* (Hewitson, 1852) **E** *Dismorphia astyocha* (Hübner, 1831) **F** *Dismorphia thermesia* (Godart, 1819) **G** *Epiphile hubneri* (Hewitson, 1861) **H** *Epiphile orea* (Hübner, 1823) **I** *Foetterleia schreineri* (Foetterle, 1902). Scale bars: 20 mm.

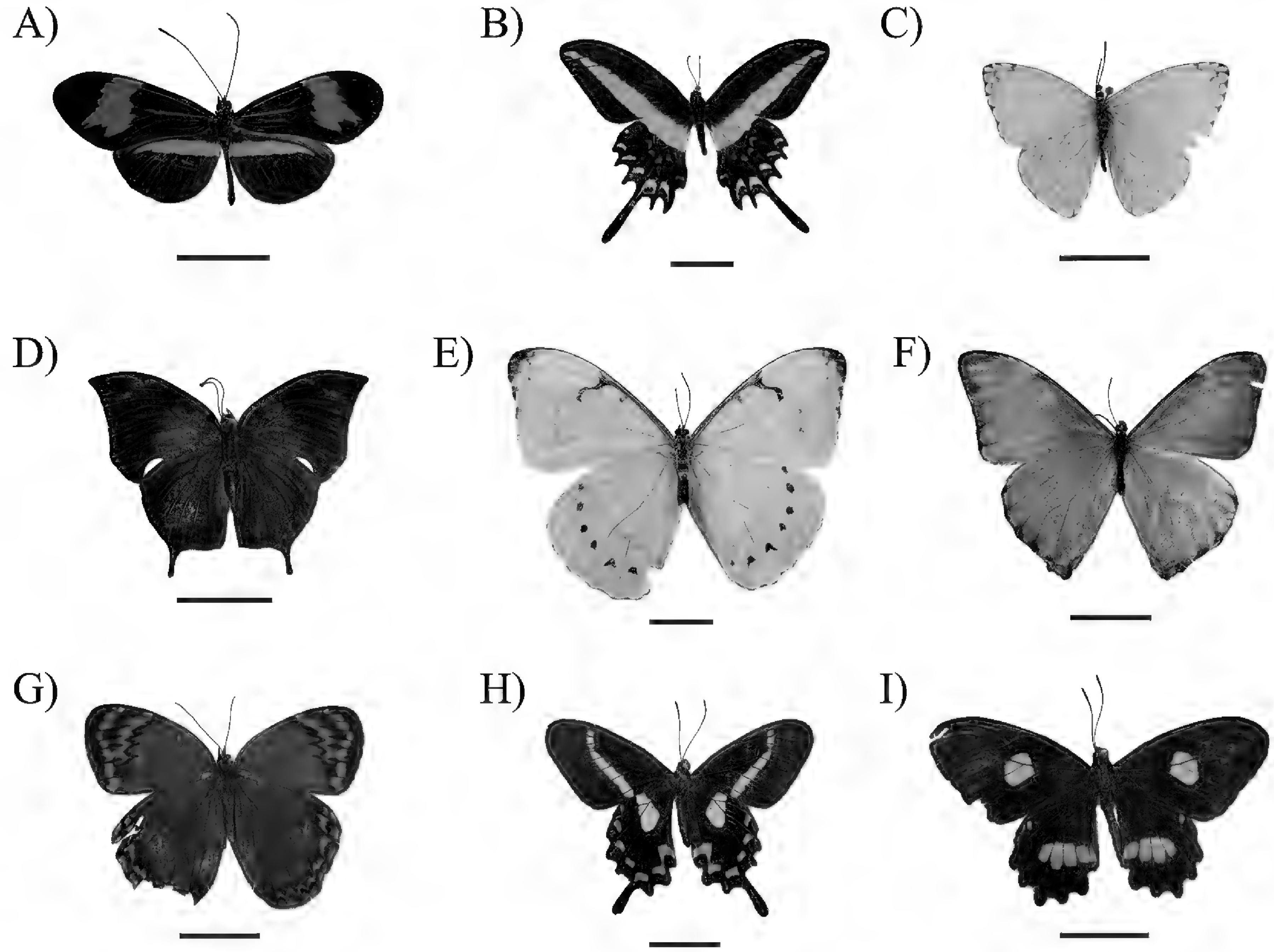

Figure 4. Butterfly species recorded in Reserva Particular do Patrimônio Natural (RPPN) Alto-Montana, Itamonte, Minas Gerais, Brazil **A** *Heliconius besckei* (Ménétriés, 1857) **B** *Heraclides hectorides* (Esper, 1794) **C** *Hesperocharis erota* (Lucas, 1852) **D** *Memphis moruus* (Fabricius, 1775) **E** *Morpho epistrophus* (Fabricius, 1796) **F** *Morpho portis* (Hübner, 1821) **G** *Opoptera syme* (Hübner, 1821) **H** *Parides agavus* (Drury, 1782) **I** *Parides anchises* (Linnaeus, 1758). Scale bars: 20 mm.

The rarefaction curve did not reveal a tendency toward stabilization (Fig. 6). However, the indices showed higher values for the 124 species sampled. The analysis performed using the Bootstrap estimator predicted a total of 143.22 species (± 10.87 SE), with 19 additional species than observed. Chao 1 predicted 153.42 (± 11.82 SE), and Jackknife 1 predicted 164.00 (± 16.29 SE) species, with 29 and 40 additional species than observed, respectively.

Discussion

Our survey showed that the butterfly community of the RPPN Alto Montana is quite diverse, especially when we take into account the size of the area in hectares. With considerable richness, our data suggested a positive potential for the RPPN Alto Montana to provide relevant information on butterfly richness for the Serra da Mantiqueira region. Although data on butterfly biodiversity is still incipient today, especially for mountainous regions (Shirai et al. 2019), works such as ours highlight the importance of fauna inventories for knowledge of communities, detection of rare and/or threatened species and identification of endemisms. We also point out the crucial role that fauna inventories can play as precursors to environmental conservation actions.

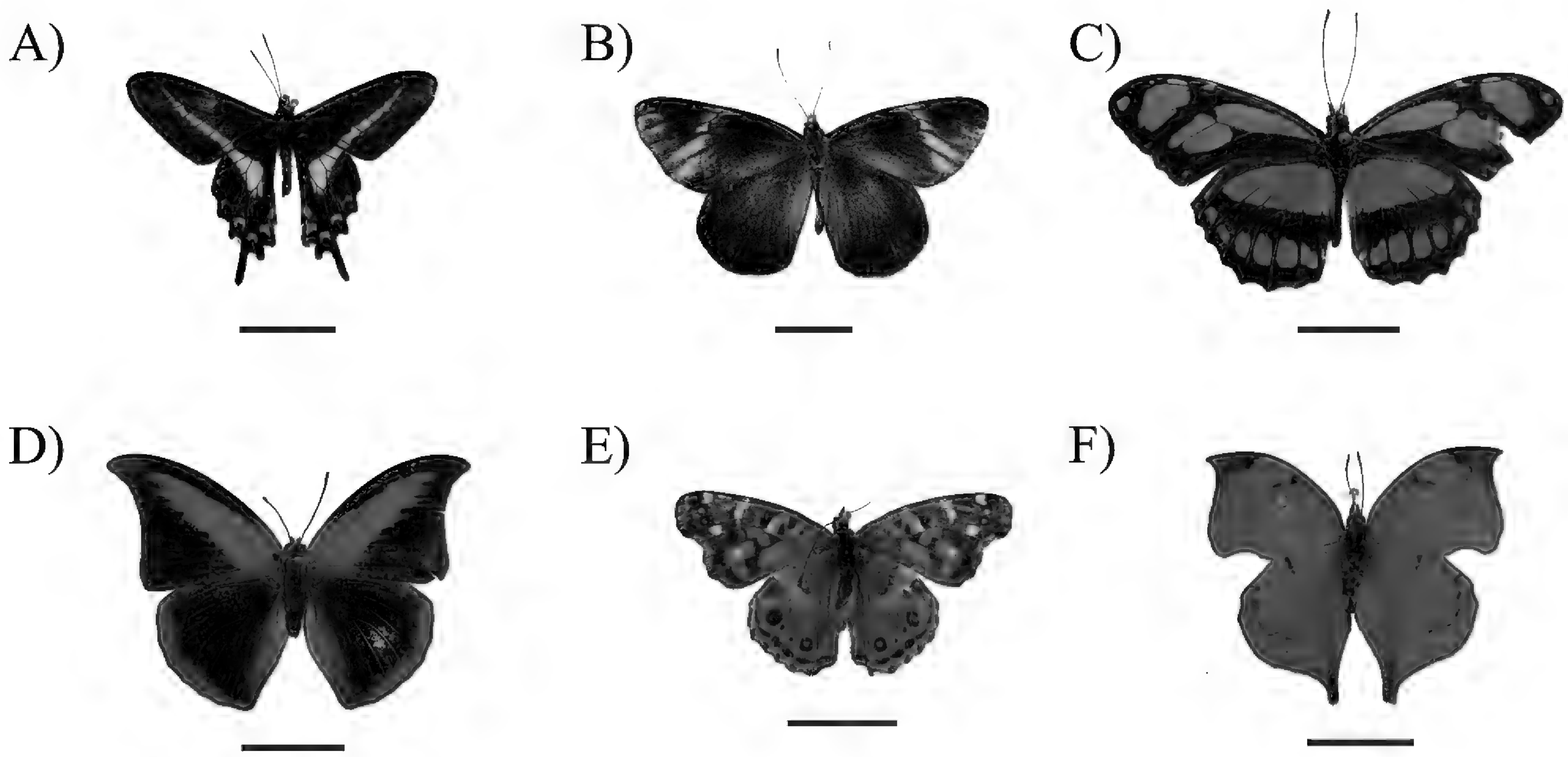

Figure 5. Butterfly species recorded in Reserva Particular do Patrimônio Natural (RPPN) Alto-Montana, Itamonte, Minas Gerais, Brazil **A** *Parides proneus* (Hübner, 1831) **B** *Pereute swainsoni* (Gray, 1832) **C** *Philaethria wernickei* (Röber, 1906) **D** *Polygrapha suprema* (Schaus, 1920) **E** *Vanessa braziliensis* (Moore, 1897) **F** *Zaretis itys* (Cramer, 1777). Scale bars: 20 mm.

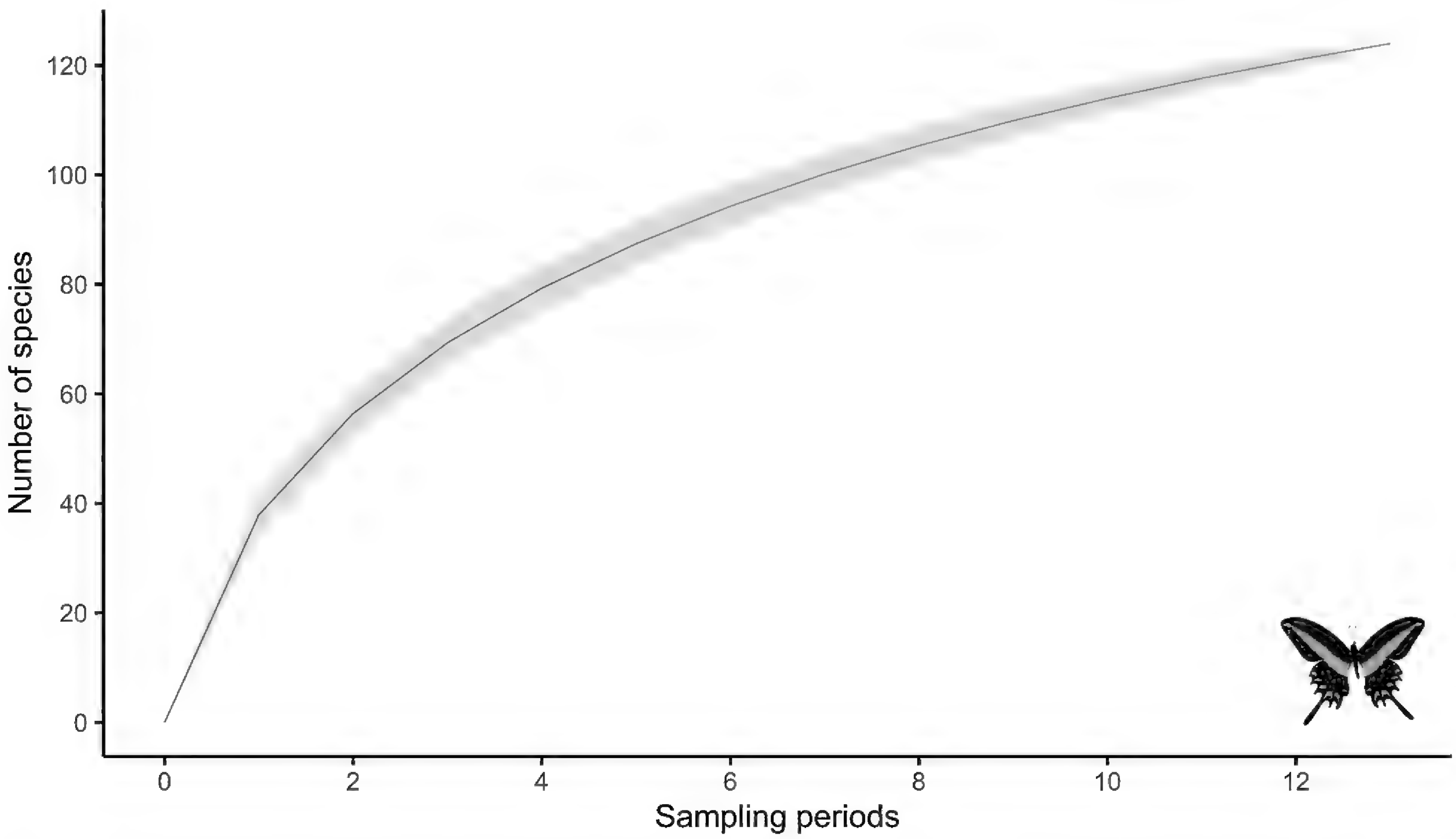

Figure 6. Rarefaction curve representing the relationship between species and sampled area. The line represents the average calculated value of the species collected at the 13 sampling periods in Reserva Particular do Patrimônio Natural (RPPN) Alto-Montana, Itamonte, Minas Gerais, Brazil. The shaded area represents the standard error (α = 0.05).

Our results showed a high richness of butterflies for a relatively small area, when we compare the extension in hectares of the RPPN Alto Montana with other areas already studied. Thus, the richness of butterfly species found in this work was higher than that observed for other mountainous regions of Brazil

(Bordin et al. 2019; Henriques et al. 2019; Gueratto et al. 2020), even presenting a considerably smaller area of 672 ha. Gueratto et al. (2020), for comparison, in Serra do Japi, SP, recorded 69 species of butterflies in 24 months of collection, a large area (35 thousand ha) when compared to the area of the RPPN Alto Montana. Our survey is also similar to studies such as Vieira et al. (2022), who worked in the Área De Proteção Ambiental Fernão Dias, south of Minas Gerais, using the same sampling methods employed here, recording 154 butterfly species, but in a larger area. Such variations in richness may occur due to differences in methodology, collection efforts, altitudinal range, and the size of sampling sites (Santos and Fernandes 2021). However, when we consider the extension in hectares of the RPPN Alto Montana, these values suggest that this conservation unit, even small, can play an important role for the conservation of butterfly communities in Mantiqueira, with potential for new butterfly records.

Comparing our study with the species lists published by Freitas et al. (2011) and Vieira et al. (2022) – both in the Serra da Mantiqueira region – we found 45 species that are also present in the Parque Nacional do Itatiaia, RJ, and 48 of the species in Área de Proteção Ambiental (APA) Fernão Dias, MG. The number of species shared between this and the studies mentioned above was expected, since these areas are located 14 and 130 km from the study area, respectively, being located in the same geographic region (Serra da Mantiqueira). However, many species were unique to each study. This exclusivity is perhaps due to the fact that there may be a certain dependence between the species and the environment in which they live. Many butterflies are sensitive to climatic and environmental variations, which could act by restricting the occurrence of species to certain regions (Chowdhury et al. 2023). The Parque Nacional do Itatiaia is a conservation unit of integral protection. The APA Fernão Dias and the RPPN Alto Montana are conservation units for sustainable use. Considering that anthropic interference in these places is different in response to the different levels of protection that each area performs, perhaps there could be a subtle difference in climate or vegetation, for example, which could explain the exclusivity of some species of butterflies for each location. Furthermore, mountains and altitude are important factors for speciation, sometimes leading many species to be restricted to a mountain peak/ridge (Montejo-Kovacevich et al. 2022).

Being a megadiverse group, studies with insects generally do not show stabilization in their rarefaction curves (Crist and Veech 2006). The Jackknife 1 estimator showed that 75.61% of the species were sampled, a higher value but very close to that found by Vieira et al. (2022) in Mantiqueira and Henriques et al. (2019) in Serra de São José, MG. For Serra do Japi, Gueratto et al. (2020) found 82% of the species according to Jackknife 1 and 63% according to Chao 1. For the RPPN Alto Montana, Chao 1 indicated 80.82% of sample sufficiency. These values are in agreement with the studies carried out for forest regions. However, taking into account the size in hectares of the studied area, these values can be a strong indication that the community of butterflies in the region is quite diverse, with more species to be registered.

The lack of stabilization in the accumulation curve may be due to the species' rarity, since almost half of the sampling species are doubletons and singletons. The classification of species recorded in singletons and doubletons showed that the evaluated butterfly community is largely represented

by rare species, 46% of the sampled richness. Rare species in the vast majority of cases are distributed in defined environmental ranges, with specific conditions (Thomson et al. 2006; Pearman and Weber 2007; Henriques et al. 2019). In the study area, replacements of tree communities along the altitudinal gradient were observed by Mariano et al. (2020). This characteristic could suggest that the occurrence of rare butterfly species in the RPPN Alto Montana would occur on microscales defined by vegetation change, restricted to small amplitude ranges. Also, the mountain climate leads to a very short window for species reproduction and activity, being a very important issue when considering sampling design (Freitas et al. 2009). For example, some Actinote species fly during short time periods, and they can be very abundant but only for one or two weeks in the summer (Freitas et al. 2009).

Nymphalidae and Hesperiidae were the most representative families in this study, as well as in similar surveys already carried out for other Atlantic Forest sampling sites in Minas Gerais (Oliveira et al. 2018; Vieira et al. 2022). This is probably due to the fact that Nymphalidae is an abundant frugivorous butterfly family found in almost all ecosystems (Brown and Freitas 2000; Bordin et al. 2019). Easily captured in attractive traps, they are generally highly representative in butterfly surveys, and is a family with high diversity in behavior and resource use variation (Bordin et al. 2019). Hesperiidae, in turn, is a large family with a predominant occurrence in open areas (Henriques et al. 2019). It has many endemic species (Warren et al. 2008), and in Brazil, its richness and abundance is concentrated mainly in high altitude areas of the Southeast region (Henriques et al. 2019). The representativeness of these families in this survey may have occurred due to some local phytophysiognomic and geographic characteristics (Mariano et al. 2020). The range of variations in resource use performed by butterflies of the Nymphalidae family, combined with the local phytophysiognomic richness (Mariano et al. 2020) may have contributed to this being the most sampled family. In the same way, active collections with sweep nets carried out in regions of borders and access trails along the altitudinal gradient or in open areas such as in Campos de Altitude, may have contributed to the capture of representatives of Hesperiidae.

Morpho portis (Hübner, 1821), *Dismorphia thermesia* (Godart, 1819), and *Foetterleia schreineri* (Foetterle, 1902) were the three most abundant species in this survey. All occur predominantly in high-altitude regions, both in reforestation areas and preserved forests (Viloria 1998; Bond-Buckup et al. 2008). *F. schreineri* is an endemic species of southeastern Brazil with known records for the Serra da Mantiqueira (Viloria 1998). *P. suprema* was the only species found in the Livro Vermelho da Fauna Brasileira Ameaçada de Extinção (Ministério do Meio Ambiente 2022) whose category is endangered (EN). Its occurrence is restricted to regions with altitudes higher than 1,500 m in Serra da Mantiqueira (ICMBio 2018) and is endemic to this region. Only five species belonging to the Papilionidae family were found on the IUCN Red List with Least Concern (LC) status (Table 1). However, little is known about the conservation of insects, including butterflies, for the entire Neotropical region (Montgomery et al. 2020), and most searches performed on the IUCN Red List for butterflies do not result in much information. Even though studies on butterflies have proliferated in

recent years, this advance is still discreet (Shirai et al. 2019). This suggests that the difficulty in knowing the conservation status of most butterfly species may be a consequence of the scarcity of studies that assess local communities. The lack of data on the species conservation listed here, added to the presence of endemisms, highlights the need for greater efforts aimed at understanding the communities and distribution patterns of the species, especially for mountainous regions, where less information is available. Our results also highlight the importance of fauna inventories as tools to support more effective conservation strategies.

Conclusion

The Atlantic Forest has suffered high losses in its biodiversity (de Lima 2020), and for the state of Minas Gerais, there are few butterfly inventories in this phytogeographic Domain (Santos et al. 2018), highlighting some of them - Andrade and Teixeira (2017), Oliveira et al. (2018), Bordin et al. (2019), Gueratto et al. (2020), and Vieira et al. (2022). In recent years, new species have been described for the Serra da Mantiqueira region such as *Actinote mantiqueira* (Freitas, Francini, Paluch & Barbosa, 2018) and *Carminda surpresa* (Barbosa, Aguiar, Rosa, Zacca & Freitas, 2020). These descriptions show the potential of Serra da Mantiqueira to contribute to the discovery of new species, expanding knowledge of biodiversity for the Atlantic Domain. Our study contributes to the knowledge of the biodiversity of tropical butterflies and reveals a high species richness within the relatively small area of the RPPN Alto Montana, in addition to providing the first inventory of butterflies for the site. Our results can support additional conservation actions in Serra da Mantiqueira due to the presence of endemic and rare species observed here, in addition to a threatened species whose occurrence is known only for the region. Finally, we also suggest with this work that there may be several species still unknown, and we encourage research in this region of Brazil.

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Additional information

Conflict of interest

The authors have declared that no competing interests exist.

Ethical statement

No ethical statement was reported.

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Author contributions

Andrêsa Garcia Andrade: Contribution in the concept and design of the study; Contribution to data collection; Contribution to butterfly identification; Contribution to manuscript preparation. Cássio Cardoso Pereira: Contribution to data analysis and interpretation; Contribution to manuscript preparation; Contribution to critical revision. Vinícius da Fontoura Sperandei: Contribution to data collection; Contribution to butterfly identification; Contribution to manuscript preparation; Contribution to critical revision. Tatiana Cornelissen: Contribution in the concept and design of the study; Contribution to data collection; Contribution to manuscript preparation; Contribution to critical revision.

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Data availability

All of the data that support the findings of this study are available in the main text.

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